OBSERVATIONS ON THE SEXUAL STATE OF VARIOUS PLANTS.*

JOHN H. SCHAFFNER
Department of Botany, Ohio State University

In recent years the writer has accumulated a considerable number of records of experiments and observations on various phases of the sexual state of plants which, although hardly extensive enough for special treatment, are nevertheless of importance in leading to a correct understanding of the nature of sexuality in the sporophytes of the higher plants. A number of the more important observations are, therefore, presented below which may give additional support to some important conclusions previously reached and published from time to time.

ACNIDA TAMARISCINA (Nutt.) Wood.

Experiments have been conducted on this diecious plant in the hope that a method for producing sex reversal might be discovered which would prove as effective as the methods used with the hemp, hop, and Arisæma. So far the effort has been only partially successful, only a few intermediates having been produced. The species is a very abundant weed in the cultivated fields of North Central Kansas where extensive observations were made in the field to determine its normal sexual expression. It seems to be normally strictly diecious. The typical staminate flower has five sepals and five stamens, while the carpellate flower has an ovulary with three prominent stigmas. The carpellate flower is described as wanting a calyx but the two minute bracts situated immediately beneath the ovulary may just as well be regarded as a reduced calyx (Fig. 1a). In either case the vegetative parts of the flower are strongly dimorphic. There are no vestiges of stamens in the carpellate flower but the staminate flower has a small vestigial gynecium without stigmas (Fig. 1b).

One carpellate intermediate plant had a large number of normal carpellate flowers in a simple spike-like inflorescence but in the middle of the inflorescence a single bisporangiate

* Papers from the Department of Botany, The Ohio State University, No. 140.

flower developed which had an ovulary with two sigmas and beside this a perfect stamen bearing pollen. (Fig. 1c). There was thus sex reversal in only one little group of cells in this entire plant which managed to develop a perfect stamen. All the remaining part of the inflorescence remained in the female state.

One staminate intermediate plant produced a flower in the leaf axil of the second leaf that had an ovulary-stamen complex with fairly well-developed stigmas (Fig. 1d). The remaining flowers were pure staminate.

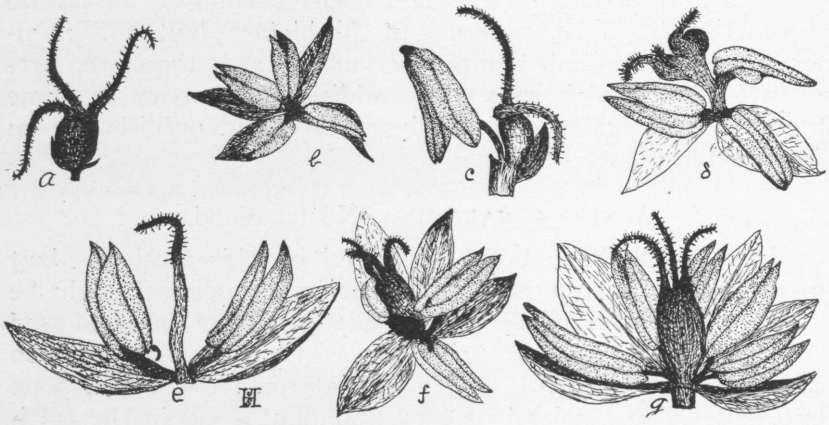

FIGURE 1

Fig. 1. Flowers of Acnida tamariscina (Nutt.) Wood. a—Typical carpellate flower from carpellate plant, showing the three stigmas, the ovulary, and the two perianth bracts or sepals below. b—A young staminate flower from a staminate plant showing the sepals, stamens, and vestigial ovulary in the center. This flower is abnormal in having only 4 stamens and 4 sepals instead of 5. c—Bisporangiate flower from a carpellate plant. This flower was in the middle of a rather long carpellate inflorescence. The perfect stamen was the only staminate structure developed on the entire plant, all the other flowers having pure female expression. d—Abnormal, intermediate flower from a staminate plant with slight expression of femaleness. Three sepals have been cut away. None of the 3 stamens are normal in appearance. The one in the stamen-carpel complex has two long microsporangia and one short one. The ovulary is only slightly developed but has three short stigmas and on one side a small neutral microsporangium. e, f, g—Flowers from an intermediate plant which had all the flowers more or less bisporangiate. The plant was probably a modified staminate plant. The flower represented by e had a rudimentary ovulary ending in a single stigma, 5 normal stamens and 5 normal sepals. f—Flower with 4 sepals, 2 normal stamens, and a fairly well developed gynecium with 3 stigmas. On the upper part of the ovulary wall, at one side, is a sessile anther with 2 microsporangia. g—Bisporangiate flower with 5 normal sepals, 5 normal stamens, and a nearly perfect gynecium with 3 stigmas.

Another staminate intermediate plant had numerous bisporangiate flowers and a number of pure staminate flowers (Figs. 1e, f, g). This intermediate plant had part of its flowers as follows:

1. A flower with 6 stamens and 6 sepals.
2. A flower with 5 stamens, an ovulary with 3 stigmas, and 5 sepals.
3. A flower with a distorted ovulary with 3 stigmas and a small anther of 2 microsporangia on its side at the top, and with 2 stamens and 4 sepals.
4. A flower with 4 stamens, an ovulary with 3 stigmas, and 5 sepals.
5. A staminate flower with 4 sepals and 4 stamens.
6. A staminate flower with 5 sepals and 5 stamens.
7. A flower with 5 sepals, 5 stamens, and a prominent vestigial ovulary with one stigma.
8. A flower with an ovulary with 3 stigmas, 5 stamens, and 5 sepals.
9. A flower with an ovulary with 5 stigmas, 2 stamens, and 5 sepals.
10. A flower with an ovulary with 3 stigmas, 3 stamens and 4 sepals.
11. A flower with an ovulary with 4 stigmas, 4 stamens and 4 sepals.
12. A flower with an ovulary with 4 stigmas, 3 stamens and 5 sepals.

The disturbance in the sexual state, caused a great confusion in the morphological expression of the flower not unlike that which occurs in the winter hemp plants, although as will be noted from the above catalog there seems to be a tendency to produce the parts in fives.

It will be seen, therefore, that although the writer has not yet discovered the proper environment for inducing considerable sex reversal in Acnida tamariscina, the short daylight period of December has apparently caused a minute percentage of reversal and this reversal is in both directions, the staminate condition to the carpellate and the carpellate to the staminate. The sexual dimorphism present was, therefore, not due to special homozygous or heterozygous sex factors, for if such had been the case reversal could at the most have taken place only in one direction. Furthermore, it is not at all a case of abnormal

chromosome complex because in that event all the flowers on a plant should have been effected somewhat similarly. The conclusion is plain that the dieciousness of Acnida is like that of the hemp caused by a reversible sexual state, any given state established at the beginning of the ontogeny being quite fixed within a certain range of environmental factors.

In Figure 1 are presented drawings of normal and intermediate flowers which will show some of the diversity of morphological expression when the sexual state has been altered in the vegetative tissues of the individual.

THALICTRUM DASYCARPUM Fisch & Lall.

In the field Thalictrum dasycarpum consists in any given season of pure carpellate plants, pure staminate plants, and a large percentage of intermediates, grading from plants which have about an equal number of staminate and capellate elements to the two extremes where a carpellate plant may have a solitary stamen and a staminate plant a single carpel, among a profusion of normal flowers. The writer asked himself the question whether the different categories of plants, pure carpellates, pure staminates, and the individuals of the various degrees of male and female expression, are fixed in their sexual nature and whether this sexual nature is due to an hereditary complex in the cells of the individual that controls the sexual expression.

A pure staminate plant and a pure carpellate plant were transferred in 1920 from a rich moist habitat to a rather dry clayey soil on the north side of the greenhouse and given no further attention except an occasional watering. In 1921, the staminate plant had some carpels. The carpellate plant was accidentally broken off at the top and did not bloom. In 1922, the staminate plant had numerous carpels and also some entirely carpellate flowers, and the carpellate plant had numerous stamens and also some flowers entirely staminate. The originally carpellate plant was decidedly more staminate the second year than carpellate.

From this simple experiment it is, therefore, plain that our notion of the fixity of the sexual state is quite wrong and that the nature of the plant may change from year to year. The cause of the different grades of sexual expression is not to be found in a study of specific hereditary constitutions. What

makes a plant pure carpellate, pure staminate, or intermediate in sexual expression in any given year or period of years is not due to any heredity compelling maleness, femaleness, or intermediateness, but is due to the fact that the heredity is of such a nature in the species as to permit the sexual expression to vary with the environment. As one finds the various types of individuals in the field, one knows that the different types must all have the same hereditary constitution which is of such a nature as to allow the sexual state to be easily changed, and the probability of an individual remaining of the same type for any length of time in a changing environment is remote. Thalictrum dasycarpum is, therefore, in strong contrast in respect to its sexual nature with a diecious species like Acnida tamariscina in which a given sexual state once established is apparently not easily changed.

ACER SACCHARINUM L.

The silver maple is a diecious species which shows intermediate individuals of varying degrees of staminateness and carpellateness and frequent reversals of the sexual state in its branches in both staminate and carpellate individuals. Often there are typical staminate flowers showing normal stamens and reduced gynecia on prevailingly carpellate plants side by side with typical carpellate flowers. Mingled with these there may be various types of intermediate flowers as follows: carpellate flower with one fruitful stamen with the other stamens reduced; carpellate flower with two fruitful stamens and the remaining ones reduced; carpellate flower with three fruitful stamens and the remaining ones reduced; flower with a normal gynecium and all the stamens enlarged and fruitful, or a completely bisporangiate flower.

Sometimes a branch on a staminate tree changes from completely staminate below to completely carpellate above with various types of more or less perfectly bisporangiate flowers on the transition zone. Branches have been found on a carpellate tree that were first carpellate, i. e., had short side branches or clusters with normal carpellate flowers, and then continued with a zone, a foot or so long, with all the flowers typically staminate, and finally by another reversal became carpellate again toward the outer end. In such cases there must be either a double reversal of the sexual state itself or of some more

primary condition leading up to the perfected sexual state. Femaleness is expressed for a time in the branch and further on maleness and still farther up the branch femaleness again becomes established.

Some trees are apparently entirely staminate or entirely carpellate at least for the season. Occasionally a tree has only a trace of the flowers of the opposite sexual state, while some appear to be about equally staminate and carpellate. Such trees may be considered to have a neutral meristem in the same condition in regard to sex as that in ordinary monecious species or species with bisporangiate flowers, while those trees that are decidedly or completely staminate or carpellate in expression may be supposed to have a physiological state already established in the meristem which under ordinary conditions invariably leads to a certain sexual expression or which under a changed environment reverses to an opposite condition leading on to the opposite sexual state.

A remarkable condition is presented by plants of the maple type of sexuality in that when the sex is reversed the conditions of development become the same as those present in trees of the opposite sexual state. A flower which has been completely reversed from the staminate condition not only has the ovulary normally developed but its stamens are reduced to vestiges although it is borne on a staminate tree, and in the same way a staminate flower on a carpellate tree has a vestigial ovulary. This peculiar phenomenon is probably not due in these cases to the diffusion of hormones from cell to cell but rather to reversals in cells or groups of cells at a given stage in the ontogeny. The vestigial structures may for a short time be in a preliminary stage leading to a given sexual state and later this state fades out in the incept to a neutral state or is even reversed to the opposite sexual state which inhibits any further development in the given line.

A sex limited character was observed in a staminate tree with some carpellate branches and some partly carpellate branches which showed the complexity of the sexual states and sexual expressions to be analyzed and interpreted. Both the normal staminate flowers and the reversed carpellate flowers had red anthers, the latter of course being vestigial, while the stigmas of the carpellate flowers were long and red and the vestigial stigmas of the staminate flowers were short and green.

ACER RUBRUM L.

The red maple is a diecious tree which seems to have less sex reversal than the silver maple. Nevertheless, it is not difficult to find staminate trees with well-developed carpellate flowers and carpellate trees with perfectly developed staminate flowers, each with its appropriate vestigial structures. The writer has found a number of such trees. The diverse nature of the sexual expression shows, as in the silver maple, that the sexual characters are not at all due to any special sexual hereditary complex of chromosomes or factors but to reversible states set up at various points in the tree which already had a distinctive trend toward a single sexual state. Segregation and aggregation of chromosomes are not involved in the sex determination or reversal.

THALICTRUM DIOICUM L.

This species of meadow-rue is rather strictly diecious. Search has been made for two seasons for intermediate individuals and five have thus far been found after a search of several hours. It is to be inferred that a small percentage of intermediate plants is produced at each blooming season. The intermediate plants were as follows: 1. A staminate plant with a single flower which had a peculiar stamen-carpel complex with a well-developed stigma. The remaining flowers were all of the normal staminate type except one which had a marginal structure half stamen and half sepal. 2. A plant exactly intermediate in sexual expression. All the flowers on this plant were bisporangiate, the carpels being mostly in the center and the stamens at the outer margin. 3. A carpellate plant with several flowers having one or more imperfect stamens. 4. A carpellate plant with a number of typical staminate flowers and some bisporangiate flowers. 5. A carpellate plant much like No. 4, but with fewer staminate flowers.

Two carpellate plants which showed some stamens were dug up in the spring of 1922, planted in pots in the greenhouse, and well fertilized with cow manure. One died but the other grew well and bloomed in September, again developing some stamens along with numerous carpellate flowers. In November the plant bloomed again, showing the same confusion of sexuality, but at the third period of blooming in January, 1923, only carpellate flowers were produced. So this individual may also appear as

a pure carpellate from time to time although it has shown its potentiality to be bisporangiate at times.*

These few plants show that the sexual state in Thalictrum dioicum ranges all the way from decidedly intermediate plants with bisporangiate flowers through every conceivable degree to strictly monosporangiate individuals. The condition is essentially the same as in Thalictrum dascycarpum with this important difference, however, that while sex reversal is very easily brought about in the latter species and there are thus always abundant intermediates present, in Thalictrum dioicum the sexual state is strongly fixed in the plant's normal environment and intermediates are quite rare. There is again no question but that sex reversal takes place in both directions, maleness to femaleness and femaleness to maleness. As is well known there are species of Thalictrum with normally bisporangiate flowers, and from this primitive condition a series of species are known grading from the irregular monecious type through dieciousness with numerous intermediates to dieciousness in which the intermediate condition is very rare. Such examples show that the attempt to explain sexual states by a simple Mendelian formula of homozygous and heterozygous hereditary constitutions falls entirely beside the mark.

Aesculus glabra Willd.

The Ohio buckeye is a monecious species with staminate and carpellate flowers commingled in the same inflorescence. Both flowers have prominent vestiges of the opposite set of sporophylls. There are also flowers that are intermediate between the two extremes, apparently of the normal bisporangiate type. The petals of the staminate flowers are more yellowsih in color than those of the carpellate flowers which have a decidedly greenish tinge and are much less open. These secondary sexual characters sometimes become quite prominent en masse when entire panicles are staminate or nearly entire panicles carpellate.

Individual trees were studied and it is evident that they grade all the way from purely staminate (observed in young plants only) to decidedly carpellate individuals. No pure carpellate plants have been observed but some have been found to have a very much larger percentage of carpellate flowers than

*At the end of this plant bloomed again with all the flowers carpellate.

staminate. Three trees were especially studied because of their apparently extreme staminateness. Some of their panicles were entirely staminate and some had a single carpellate flower; others had two or more carpellate flowers with well-developed gynecia. A young tree, as stated above, probably in its first or second blooming season was found to be purely staminate in sexual expression. Such pure or decidedly staminate trees can sometimes be discovered through the bright appearance of their flower clusters. In this young tree the flower clusters were compact and very showy with bright yellow petals, because of the extreme condition of the male state.

It is possible that the sexual state changes from time to time because of changing environment or the progress of senility, but no definite observations have been made along this line except that some trees appear to be much more fruitful in some years than in others, which might, of course, be due to other causes than the relative amount or degree of maleness or femaleness expressed for the giving seasons. From these observations, the buckeye tree is found to be a monecious species which has individuals ranging, in any given year, in sexual expression, from pure staminate to decidedly carpellate. This range of the sexual condition from pure staminate to decidedly carpellate but not to the complete carpellate condition recalls a somewhat similar state of affairs in Arisæma dracontium (L.) Schott.

GENERAL DISCUSSION.

Although the careful study of the sexual states of monecious and diecious trees is attended with some difficulty, such studies would assist materially to a correct understanding of the nature of sex and the way in which the sexual state is developed in the individual or a part of the individual. The higher conifers would apparently also be an especially fruitful region for study as well as the interesting order of Sapindales to which buckeyes and maples belong.

The study of monecious and diecious species has shown that moneciousness and dieciousness are but extremes of a common series of sexual phenomena and are of varying degrees of intensity and extent. The intensity of the sexual state is independent of the monecious or diecious condition. In either state a species may be decidedly constant or decidedly unstable. The diecious species may have the given sexual state so slightly developed that even under the ordinary environment reversals take place

quite frequently; the states may be so fixed that reversals occur only in the presence of unusual ecological conditions; or they may be so intensely developed that so far no intermediates are known to have been produced, either in the natural environment or when the plant is growing under extremely different ecological conditions. Similarly, monecious species may be so constituted that there is little variation between individuals in respect to their sexual expression or they may have the given neutral or sexual state so slightly established in the cells of the individual, that determinations or reversals do not follow any very regular course in the ontogenetic development, in consequence of which the type of expression varies decidedly from one individual to the next. The lowest type of monecious species is that in which the one or the other sexual state is established permanently at the base of the floral axis, resulting in monosporangiate flowers. In the more extreme types, the time of determination of the sexual state is shifted back to an earlier axis thus producing entire inflorescences with one type of flower or entire branching systems involving numerous inflorescences or even considerable parts of the vegetative structures. Besides these there is the very common type in which sex reversal takes place at a given level or zone in the inflorescence itself simulating the sexual zonation to be observed in the ordinary bisporangiate flower.

The diecious condition is presumably brought about by the establishment of an hereditary constitution in which sex determination is coincident with fertilization in the ordinary environment in which the species lives. This shifting of the time of determination has nothing to do with the relative intensity or permanence of the sexual state set up; for as the writer has ascertained, both from observation and experiment, sex reversal and changes in the sexual state can be brought about as easily, if not more so, in the diecious sporophyte individual as in the monecious. If the sex is determined or reversed at the time of fertilization, i. e., at the time of the fusion of the male and female nuclei, the cause and the mode of determination must, nevertheless, be fundamentally the same as when the determination or reversal takes place in the vegetative incept of the flower bud or sporophyll. *Before we could assume that a new functional activity, a new chemistry or physics, was at work, we would need some very strong evidence for the assumption. This observation applies to unisexual individuals with allosomes as well as to those without.*

Bisporangiateness, moneciousness, and dieciousness, therefore, fall into a progressive series of many stages in respect to the intensity and fixity of the sexual state, all depending on a fundamentally similar physiological activity, the more prominent stages being as follows:

1. Typical bisporangiate species.
2. Bisporangiate species with the development of some monosporangiate flowers, either staminate or carpellate, or both.
3. Monecious species with some bisporangiate flowers.
4. Monecious species normally without bisporangiate flowers.
5. Monecious species which produce individuals of a greater or less degree of staminateness or carpellateness in the individual, together with occasional individuals purely staminate or purely carpellate or with both these types occasionally present.
6. Diecious species with numerous intermediates of all degrees and with sex reversal easily accomplished.
7. Diecious species with few intermediates and with apparently little sex reversal.
8. (?) Diecious species with no intermediates and no sex reversal at least under normal conditions.

Received for publication February 1, 1923.